

PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION  
International Bureau



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

|  |           |  |
|--|-----------|--|
| <b>(51) International Patent Classification <sup>7</sup> :</b><br><b>A61K 51/00</b>  | <b>A1</b> | <b>(11) International Publication Number:</b> <b>WO 00/43045</b><br><b>(43) International Publication Date:</b> 27 July 2000 (27.07.00)  |
| <b>(21) International Application Number:</b> PCT/US00/01815<br><b>(22) International Filing Date:</b> 25 January 2000 (25.01.00)<br><b>(30) Priority Data:</b><br>09/236,247 25 January 1999 (25.01.99) US<br><b>(71) Applicant:</b> PHOTOGEN, INC. [US/US]; 7327 Oak Ridge Highway, Knoxville, TN 37931 (US).<br><b>(72) Inventors:</b> WACHTER, Eric; 138 Bay Path Drive, Oak Ridge, TN 37830 (US). SMOLIK, John; 119 Tanasi Court, Loudon, TN 37774 (US). DEES, H., Craig; Apartment 1517, 1006 Wyndham Way, Knoxville, TN 37923 (US).<br><b>(74) Agents:</b> MANZO, Edward et al.; Cook, Alex, McFarron, Manzo, Cummings & Mehler, Ltd., Suite 2850, 200 W. Adams Street, Chicago, IL 60606 (US). |           | <b>(81) Designated States:</b> AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).<br><br><b>Published</b><br><i>With international search report.</i><br><i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i> |
| <b>(54) Title:</b> METHOD FOR IMPROVED RADIATION THERAPY<br><br><b>(57) Abstract</b><br><br>A method is disclosed for treating a selected volume of tissue which method includes distributing a radiosensitizer and a plurality of ionizing radiation sources substantially within the volume of tissue to produce treatment zones that are generally uniformly distributed throughout the volume of tissue. An agent is also disclosed for treating such tissue, wherein the agent includes a radiosensitizer and an ionizing radiation source used in conjunction to define an injectable treatment agent.   |           |  |

28

**FOR THE PURPOSES OF INFORMATION ONLY**

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

|    |                          |    |  |    |  |    |                          |
|----|--------------------------|----|--|----|--|----|--------------------------|
| AL | Albania                  | ES | Spain                                    | LS | Lesotho                                      | SI | Slovenia                 |
| AM | Armenia                  | FI | Finland                                  | LT | Lithuania                                    | SK | Slovakia                 |
| AT | Austria                  | FR | France                                   | LU | Luxembourg                                   | SN | Senegal                  |
| AU | Australia                | GA | Gabon                                    | LV | Latvia                                       | SZ | Swaziland                |
| AZ | Azerbaijan               | GB | United Kingdom                           | MC | Monaco                                       | TD | Chad                     |
| BA | Bosnia and Herzegovina   | GE | Georgia                                  | MD | Republic of Moldova                          | TG | Togo                     |
| BB | Barbados                 | GH | Ghana                                    | MG | Madagascar                                   | TJ | Tajikistan               |
| BE | Belgium                  | GN | Guinea                                   | MK | The former Yugoslav<br>Republic of Macedonia | TM | Turkmenistan             |
| BF | Burkina Faso             | GR | Greece                                   | ML | Mali   | TR | Turkey                   |
| BG | Bulgaria                 | HU | Hungary                                  | MN | Mongolia                                     | TT | Trinidad and Tobago      |
| BJ | Benin                    | IE | Ireland                                  | MR | Mauritania                                   | UA | Ukraine                  |
| BR | Brazil                   | IL | Israel                                   | MW | Malawi                                       | UG | Uganda                   |
| BY | Belarus                  | IS | Iceland                                  | MX | Mexico                                       | US | United States of America |
| CA | Canada                   | IT | Italy                                    | NE | Niger  | UZ | Uzbekistan               |
| CF | Central African Republic | JP | Japan                                    | NL | Netherlands                                  | VN | Viet Nam                 |
| CG | Congo                    | KE | Kenya                                    | NO | Norway                                       | YU | Yugoslavia               |
| CH | Switzerland              | KG | Kyrgyzstan                               | NZ | New Zealand                                  | ZW | Zimbabwe                 |
| CI | Côte d'Ivoire            | KP | Democratic People's<br>Republic of Korea | PL | Poland                                       |    |                          |
| CM | Cameroon                 | KR | Republic of Korea                        | PT | Portugal                                     |    |                          |
| CN | China                    | KZ | Kazakhstan                               | RO | Romania                                      |    |                          |
| CU | Cuba                     | LC | Saint Lucia                              | RU | Russian Federation                           |    |                          |
| CZ | Czech Republic           | LI | Liechtenstein                            | SD | Sudan  |    |                          |
| DE | Germany                  | LK | Sri Lanka                                | SE | Sweden                                       |    |                          |
| DK | Denmark                  | LR | Liberia                                  | SG | Singapore                                    |    |                          |
| EE | Estonia                  |    |  |    |  |    |                          |

## METHOD FOR IMPROVED RADIATION THERAPY

### BACKGROUND OF THE INVENTION

5 The present invention relates to methods and compositions for radiation treatment of a selected volume of tissue, such as the treatment of abnormal cellular proliferation or malignant tissue. More specifically, the present invention relates to a novel method that involves distributing a radiosensitizer and a plurality of ionizing radiation sources substantially within the volume of tissue to produce treatment zones that are generally uniformly distributed throughout the volume of tissue. The present invention also  
10 particularly relates to a novel agent for treating such tissue wherein the agent includes a radiosensitizer and an ionizing radiation source in conjunction to define an injectable treatment agent.

It is well known that abnormal cellular proliferation occurring in cancer, for example, is often treated using ionizing radiation in a process known as radiation therapy.  
15 In radiation therapy (which typically but not necessarily uses electromagnetic radiation with energies of 1 keV or higher), radiation may be applied from an external source or through introduction of a radioactive source inside tissue to be treated. One of the well-known drawbacks of radiation therapy is collateral damage to healthy tissue surrounding the primary treatment site. Accordingly, a major challenge in radiation therapy is selective  
20 delivery of a therapeutic dose of radiation to the desired tissue, such as that in a cancerous tumor. One technique for localizing the effect of the radiation is to use radiodense or radiopaque radiosensitizing agents in conjunction with applied radiation. These agents enhance the effect of the radiation on the tissue treated with the sensitizer and have in the past, yielded dose enhancement (DE) in tumors, when such agents are introduced in the  
25 tissue to be treated.

The use of sensitizers, however, is only an advance, not the complete answer in radiation therapy. There are a number of significant problems in prior methods when using such an approach, especially in the uniform treatment of a desired tissue or treatment volume. For example, after administering the radiopaque radiosensitizing agent,  
30 uniform delivery of a therapeutic radiation dose to the entire treatment volume is often thwarted by portions of the treatment volume being shielded by the radiosensitizer.

More specifically, when using external radiation, where radiation is delivered from a source outside the body, the radiosensitizer in the treated tissue closest to the radiation

source may tend to shield or block the more distal tissue from the ionizing radiation. As a result, only part of the tissue to be treated receives an effective radiation dose.

Similarly, when a confined radiation source, such as a radioactive needle or the tip of a wire or ribbon, is introduced directly into or into the proximity of the tissue to be treated (commonly called brachytherapy), the radiosensitizer in proximity to the radiation source may act to shield the remainder of the tissue from the desired dose. The result is less than uniform treatment of the tissue volume, such as a tumor, in question.

Accordingly, it is a general object of the present invention to improve the uniformity of radiation treatment for selected volumes of tissue, such as tumors and the like.

Another object of the present invention is to provide methods and agents for radiation treatment of tissue that help provide more uniform treatment of desired volume of tissue.

### SUMMARY OF THE INVENTION

The present invention is directed to a method for treating a selected volume of tissue. The method comprises the steps of distributing a radiosensitizer within the volume of tissue; and distributing a plurality of ionizing radiation sources within the volume of tissue with each radiation source producing a radiation treatment zone. The radiation sources are distributed within the volume of tissue such that the treatment zones from the plurality of radiation sources are generally distributed throughout the volume of tissue and substantially all of the tissue in the selected volume is within at least one treatment zone.

In a preferred embodiment of the present invention, the radiosensitizer and/or the radiation sources are substantially uniformly distributed within the selected volume to afford a more uniform treatment to the tissue within the selected volume. In accordance with another aspect of the present invention, each of the treatment zones may overlap at least one other treatment zone to better assure effective treatment of the tissue in the selected volume.

In accordance with other aspects of the present invention, the distributing of a radiosensitizer and the distributing of ionizing radiation sources may be carried out sequentially or simultaneously, by injecting them together, directly into or into proximity to the selected volume of tissue or, in another aspect of the present invention, the distributing of a radiosensitizer and the distributing of ionizing radiation sources may be carried out by sequentially administering the radiosensitizer and the ionizing radiation sources.

To avoid undue effect on healthy or other tissue outside the selected volume, in accordance with another aspect of the present invention, the treatment zones of the radiation source preferably do not extend substantially beyond the selected volume of tissue.

The present invention, in accordance with another aspect, is also directed to an agent for treatment of tissue. Preferably, the agent comprises a radiosensitizer component and an ionizing radiation source component, with the radio sensitizer component and radiation source component being combined to define an injectable treatment agent. The agent may be of any suitable form, but in preferred aspects of the present invention is a liquid or gel. Also, the radio sensitizer and radiation source may be conjugated to define an injectable agent.

These are but a few of the many aspects of the present invention that are set forth in the appended claims and described in the following detailed description of the invention.

### **BRIEF DESCRIPTION OF THE DRAWINGS**

5 Fig. 1a illustrates an example of shielding of the interior and distal portions of a treatment region using prior treatment methods;

Fig. 1b illustrates an example of shielding of the distal portions of a treatment region using an internal source with prior treatment methods;

10 Fig. 2a illustrates the effect of a single tumor-dispersible radioactive material in conjunction with a tumor-specific radiosensitizer in a desired treatment volume;

Fig. 2b illustrates a preferred embodiment of the present invention wherein multiple entities of tumor-dispersible radioactive material are present in the treatment volume of Fig. 2a;

15 Fig. 3a illustrates the linkage of a radiosensitizer moiety and a radioactive moiety to produce a sensitizer-radioconjugate agent, in accordance with an embodiment of the present invention;

20 Fig. 3b illustrates a conjugate radiosensitizer moiety attached to a targeting moiety to produce a sensitizer conjugate, and a conjugate radioactive moiety attached to a targeting moiety to produce a radioconjugate agent, in accordance with an embodiment of the present invention; and

Fig. 3c illustrates a radiosensitizer moiety and a radioactive moiety in a delivery vehicle, in accordance with an embodiment of the present invention.

### **DETAILED DESCRIPTION OF THE PRESENTLY PREFERRED EMBODIMENTS**

25 Figures 1a and 1b illustrate radiation shielding or blocking problems in the prior art. In particular, Figure 1a shows an example of shielding10 of interior and distal portions of a treatment volume. In this example, a radiopaque sensitizer 16 has been distributed throughout the treatment volume 18, such as a cancerous or malignant tumor. When radiation 12 is applied using an external source 14, the sensitizer closest (20) to the  
30 external source 14 substantially absorbs a major portion of the radiation 12 before it can reach sensitizer located distal (22) to the external source. As a result, the interior and distal portions of the treatment volume 18 are shielded from the radiation 12, and the efficiency of activation of the distal sensitizer 22, located beyond a certain radiation

penetration distance 24, is severely reduced. This self-shielding yields an uneven treatment, where most or all activation occurs in a sub-volume 26 which is primarily confined to that portion of the treatment volume 18 proximal to the source 14 (i.e., at a source-target separation within the radiation penetration distance 24). Even with delivery  
5 along multiple paths, such shielding occurs at interior portions of the treatment volume, reducing efficacy of the overall treatment regimen.

Another example of shielding 30 is shown in Fig. 1b, where a contained, internal source is used, such as for example in brachytherapy. As shown in Figure 1b, a radiopaque sensitizer 16 is distributed in a substantially uniform fashion throughout the  
10 treatment volume 18. Shielding of distal portions of a treatment volume 18 occurs when radiation 12 is applied using an internal source 32. The sensitizer present proximal to the internal source 34 substantially absorbs a major portion of the radiation 12 before it can reach sensitizer present distal to the internal source 36, thereby shielding and reducing  
15 efficiency of activation of such distal sensitizer 36 located beyond a certain effective radiation penetration distance 24. This self-shielding yields an uneven treatment, where most or all activation occurs in a sub-volume 38 which is primarily confined to that portion of the treatment volume 18 proximal to the source 32 (i.e., at a source-target separation within the radiation effective penetration distance 24).

In accordance with the present invention, and in contrast to the prior art, a method  
20 is provided for more uniform radiation treatment of a selected volume of tissue, such as a malignant mass or tumor.

As illustrated in Figures 2a and 2b, the method of the present invention, in general, includes distributing a radiosensitizer 44 within the volume 46 of tissue to be treated, and distributing a plurality of ionizing radiation sources 42 within the volume. Each radiation  
25 source 42 emits radiation in all directions which has an effective radius, before being unduly shielded, blocked, or attenuated by the sensitizer 44, tissue or other material in the tissue, to define a treatment zone 56 within which the radiation provides the desired dose enhancement effect acting in conjunction with the radiosensitizer. The radiation sources are distributed throughout the tissue volume so that substantially all of the tissue in the  
30 treatment volume lies within one or more treatment zones. Preferably the radiosensitizer and radiation sources are distributed substantially uniformly throughout the selected volume of tissue to better provide generally uniform and effective radioactive therapy to the selected tissue volume.

In a preferred embodiment of the present invention, multiple entities of tumor-dispersible radioactive material 42 in the form of multiple radioactive particles or nanoparticles are located within the treatment volume, as illustrated in Fig. 2b. By increasing the concentration of such entities of tumor-dispersible radioactive material 42 in the treatment volume 46, such that the material's multiple localized treatment zones 56 overlap, treatment 64 effected within the entire desired treatment volume 46. The tumor-dispersible radioactive material serves, once distributed, preferably substantially uniformly, throughout the tumor (where such distribution from one or more the primary administrative sites occur via natural dispersion, dilution, or other substantially physician-passive distribution mechanisms), as a uniformly dispersed source that produces a substantially uniform radiation throughout the tumor volume. Further, use of such distribution mechanisms delivers minimal effective radiation dose to healthy tissues outside the treatment volume. Such uniform radiation results in a more uniform activation of said radiosensitizers throughout the tumor volume (owing between radiation source and target, namely tumor-dispersible radioactive material and radiosensitizer as discussed *infra*), and thereby yields a more simple and effective means for delivering a therapeutic dose of radiation therapy to the entire tumor. This mitigates the effects of shielding of any given entity 42, greatly simplifying dose calculation and delivery. Since tumor-specific radiosensitizer 44 is substantially present only within the desired treatment volume 46 as explained *supra*, the existence of a portion 66 of any radiation interaction volume 56 extending beyond the desired treatment volume 46 will not produce significant collateral damage in such irradiated adjacent volumes 68. Also described *supra*, such delivery of multiple entities of tumor-dispersible radioactive material 42 can be easily effected through intratumoral injection of a solution of such material or other similar administrative techniques, such as proximal venous or lymphatic injection.

The treatment volume or selected volume of tissue may be any particular tissue that is selected for treatment. Typically, but not exclusively, the tissue to be treated will be tissue in which proliferative cell growth is occurring and which can be treated by ionizing radiation. Although such cell proliferation may be benign, it is anticipated that the present invention will find greater application in treating malignant tissue in the form of a tumor, such as for example a cancerous tumor, or other malignancy, or a mass of diseased tissue, such as a cyst, polyp or abscess.



The present invention is not limited to the specific radiosensitizer employed. Specific examples of radiosensitizers that may be used include the various radiodense halogenated xanthenes and their derivatives, such as Rose Bengal and its various derivatives, Pholixine B and its various derivatives, Erythrosin B and its various derivatives, Eosin Y and its various derivatives, along with various other highly brominated or iodinated halogenated xanthenes and their various derivatives, such as 4,5,6,7-Tetrabromoerythrosin; various x-ray contrast agents, such as Omnipaque™ (iohexol), Omniscan™ (gadodiamide), WIN 8883 (diatrizoic acid), and iodamide, and lipiodol, along with those agents containing various radiodense elements or moieties, such as iodine, bromine, chlorine, barium, bismuth, boron, gold, silver, platinum, iron, gadolinium, dysprosium, and tantalum; iododeoxyuridine and bromodeoxyuridine and related agents; various halogenated nucleotides and DNA ligands and intercalators, including various substituted acridine and imidazole based agents; various nitroimidazoles and other related bioreductive agents; misonidazole and related agents; etanidazole and related agents; pimondazole and related piperidine-derivatives; aziridines and related agents; cyclophosphamide and related agents; nitrosoureas and related agents; L-phenylalanine mustard and related agents; cis-platinum compounds and related agents; and doxorubicin and related agents. Preferably, the present invention uses a tumor-specific radiosensitizer, such as a halogenated xanthene. More preferably, the tumor-specific radiosensitizer is Rose Bengal.

In general, these radiosensitizers can be introduced into the treatment volume by systemic administration, such as intravenous administration, direct injection, or similar conventional techniques. More preferably, such administration includes direct injection or other administrative techniques into or proximal to the desired treatment volume. Use of such administrative techniques with various radiosensitizers, such as those described above can lead to a localized retention of a therapeutically useful level of such radiosensitizers within the desired treatment volume for a period of several hours to several weeks. As a result, the radiosensitizer becomes substantially uniformly distributed throughout the desired treatment volume, through dispersion, dissolution, or other passive equilibration or concentration processes, including preferential uptake.

The method of the present invention further includes the step of distributing a plurality of ionizing radiation sources. The present invention is not limited to the number of ionizing radiation sources, but contemplates the use of more than one ionizing or other

high energy radiation sources. An example of such a source, for use in the present invention, is a tumor-dispensible radioactive material, as discussed *intra*. The present invention, however, is not limited to such radioactive material, as tumor dispersible radioactive material and radioactive moieties mentioned, such as those discussed *infra*, and other similar materials, can also be used. Preferably, the one or more sources is located within the treatment volume. Further, as explained below, it is preferred that a plurality of such radiation sources be distributed within the treatment volume.

More specifically, Figure 2a illustrates the effect of a single radiation source, for example a tumor-dispersible radioactive material 42, located in a treatment volume wherein a quantity of tumor-specific radiosensitizer 44, such as for example that described supra, is substantially uniformly distributed and contained within the treatment volume 46. "Tumor-dispersible" means readily dispersible within a treatment volume, for example a tumor. Typically, a tumor dispersible radioactive material may be a liquid, gel or other dispersible form or formulation of a radioactive material made up of a fine particulate or colloidal suspension, dissolved or otherwise solubilized or dispersible in form, of a radioactive material that is stable upon injection, or other administrative technique, into a patient's body. Preferably, the tumor-dispersible radioactive material is injected, or other administered by commonly known methods such as intravenous drip or injection, directly into or proximal to a treatment volume, such as a cancerous tumor.

Examples of such tumor-dispersible radioactive materials include radioisotopes that are attached to or encompassed in organic or inorganic microspheres, micelles, or nanoparticles, or are solubilized using chelates or other organic or inorganic agents. It is well known in that art that such materials, when locally administered into or proximal to diseased tissue, such as a cancerous tumor, can exhibit prolonged retention in said tissue, with biological half-lives ranging from several hours to several weeks.

Isotropic radiation 48 emitted by the tumor-dispersible radioactive material 42 activates a portion of the tumor-specific radiosensitizer 50 that is present within a radiation interaction volume 52 (defined by the penetration distance of such radiation 48 within the tumor/radiosensitizer environment) to produce a localized treatment zone 56. Since any radiation 58 that reaches beyond this localized treatment zone 56 is of insufficient intensity to substantially activate radiosensitizer 60 present outside this localized treatment zone 56, the localized treatment zone 56 is surrounded by a non-therapeutic or non-treated volume 62. Thus, in most cases a single entity of tumor-

dispersible radioactive material 42, such as a single radioactive particle or nanoparticle, will be insufficient for complete treatment of the entire desired treatment volume 46.

The present invention overcomes this apparent short coming through the use of a plurality of sources within the treatment volume. Preferably, radiation sources are distributed substantially uniform within the volume of tissue such that the treatment zones for each source are contiguous or overlap, and there are sufficient number of radiation sources so that substantially all of the tissue volume to be treated lies within at least one treatment zone. The radioactive sources also may be distributed within the treatment volume so that the areas of tissue to be treated that reside in the outer reaches of a treatment zone, such as for example, the outer third of the zone, lie within two or more zones to better assure sufficient radiation treatment. In other words, the treatment zones overlap, and some of the tissue volume lies within two or more zones.

Preferably, the radiosensitizer and tumor-dispersible radioactive material (such material also being known as a radiopharmaceutical agent) are administered to the patient (directly into or proximal to the tumor or other diseased tissue) by (1) simple injection or other similar delivery technique of a mixed solution of each, or (2) by sequential administration of each (by simple injection or other similar delivery technique). It is further preferred that such administration be performed using a concentration and volume of the radiosensitizer material selected so as to deliver a dose of more than about 1 nanogram radiosensitizer per kilogram diseased tissue (i.e.  $\geq \text{ng/kg}$ ) but not more than about 10 gram radiosensitizer per kilogram diseased tissue (i.e.  $\leq 10 \text{ g/kg}$ ), such does being selected so as to produce sufficient localized cytotoxicity in said diseased tissue upon irradiation while avoiding induction of non-specific or systemic toxicity or cytotoxicity from the material alone. Moreover, it is further preferred that such administration be performed using a concentration and volume of said radioactive material selected so as to deliver a dose to the tissue within a selected volume of more than about 1 milliGray (i.e.  $\geq 1 \text{ mGy}$ ) but not more than about 1000 Gray (i.e.  $\leq 1000 \text{ Gy}$ ), with a preferred dose in the range from about 0.1 Gy to 100 Gy. The dose is selected so as to produce sufficient localized activation of the radiosensitizer while avoiding induction of non-specific or systemic damage from the radioactive material alone.

Concerted delivery of materials may be facilitated by utilizing radiosensitizer and tumor-dispersible radioactive materials having similar dimensional and chemical or biological properties (such as hydrophilicity or lipophilicity), since dispersion of such

agents will tend to occur in a similar fashion, resulting in substantially uniform distribution of both radiosensitizer and tumor-dispersible radioactive material at the treatment site.

5 In contrast with prior methods, such an approach has a number of distinct advantages. Since administration of radiosensitizer and radioactive material can be performed via injection or other simple administrative techniques, the complexity of the procedure and required apparatus are greatly reduced relative to that required for application of radiation using external sources or implantable sources. Such administration is possible using a single step where radiosensitizer and radioactive material are administered together in a mixed formulation. Alternatively, a simple two or more step administration procedure may be used. In this procedure, materials are administered in a staged or separate manner, for example, to allow one component to reach uniform distribution prior to administration of the second component, or where multiple doses of one or more component are required to maintain a therapeutic level, for example to compensate for radioactive decay or other loss of one or more component from the treatment volume. Further, if radiosensitizers and radioactive materials that are capable of quick decay or natural clearance from the body are utilized, the complexity of follow-up procedures are also greatly reduced, since upon destruction of the tumor such agents will either decompose or be otherwise cleared from the tumor site and patient by natural processes. Hence, unlike brachytherapy, no residual material will be left at the site (such as brachytherapy needles that might require surgical removal).

20 In some specialized cases, it may be necessary to more carefully couple the delivery of radiosensitizer and radiopharmaceutical agent in a more refined manner. In accordance with a further embodiment of the present invention, methods and agents for achieving such coupling are illustrated in Figures 3a, 3b, and 3c. In general, the agent includes a radiosensitizer component and an ionizing radiation source component combined to form an injectable treatment agent.

25 More specifically, Figure 3a illustrates a linkage of one or more radiosensitizer moiety 70, such as for example one of the radiosensitizers discussed supra, and one or more radioactive moiety 72, using for example covalent attachment or other chemical or physical mechanisms 74, to produce a sensitizer-radioconjugate agent 76. Such covalent linkage may consist, for example, of a covalent bond between a ligand-complexed radioactive moiety and an organic radiosensitizer. Such a conjugate agent 76 assures that

radiosensitizer and radiation source are delivered together in the proper stoichiometry, in a manner similar to that discussed above. Preferably, multiple entities of such conjugate agents are administered to the treatment volume or region, as discussed above.

The present invention is not limited to a specific radioactive moiety. Specific examples of radioactive moieties that can be used in the present invention include various radioisotopes and chemical derivatives of such radioisotopes, including those of aluminum, americium, cobalt, copper, gallium, gold, indium, iodine, iridium, manganese, phosphorus, radium, rhenium, rhodium, ruthenium, sulfur, technetium, thallium, yttrium, and of other radioactive elements, compounds, or materials capable of producing  $\alpha$ -,  $\beta$ -,  $\gamma$ -, x-ray, or other high energy or ionizing radiation.

An alternate embodiment, illustrated in Figure 3b, uses (a) a radiosensitizer moiety 70, such as for example those discussed supra, attached to a chemical or biological targeting moiety 78 to produce a sensitizer conjugate 80, and (b) a radioactive moiety 72, such as for example those discussed above, attached to a similar chemical or biological targeting moiety 78 to produce a radioconjugate 82. Delivery of the sensitizer conjugate 80 and radioconjugate 82, in a manner similar to that discussed above, for example via injection, either together or sequentially, facilitate uniform combined delivery of both elements to the desired treatment site, based on specificity of the targeting moieties 78 for a disease site.

A further alternate embodiment of the present invention, illustrated in Figure 3c, includes one or more radiosensitizer moiety 70 and one or more radioactive moiety 72, such as for example the moieties discussed above, in a delivery vehicle 82. Examples of such delivery vehicles include a micelle, liposome, or nanoparticle, formed using methods commonly available in the art. Such an encapsulated agent 84 can be designed to deliver its contents 70 and 72 to a specific treatment area or cellular structure, such as cell membranes, so as to further augment the efficacy of dose enhancement, as described in U.S. application no. 09/216,787 which is incorporated herein by reference.

Specific examples of the chemical or biological targeting moieties, as described in reference to Figure 3a and 3b, include DNA, RNA, amino acids, proteins, antibodies, ligands, haptens, carbohydrate receptors or complexing agents, protein receptors or complexing agents, lipid receptors or complexing agents, chelators, encapsulating vehicles, nanoparticles, short-or-long-chain aliphatic or aromatic hydrocarbons, including

those containing aldehydes, ketones, alcohols, esters, amides, amines, nitriles, azides, or other hydrophilic or hydrophobic moieties.

Accordingly, it is a preferred embodiment of the present invention to combine delivery of an efficient radiosensitizer, such as for example Rose Bengal, and a tumor-dispersible radioactive material, such as for example rhenium-188 ( $^{188}\text{Re}$ ), as an injectable mixture into diseased tissue, such as a cancerous tumor.  $^{188}\text{Re}$  is a generator-produced gamma emitter (155 keV) that is readily packaged in nanoparticulate form, with a biological half-life of approximately 1 week. Such gamma energy is readily absorbed by iodine atoms contained in Rose Bengal, which transform such energy to a therapeutically active form (such as Auger electrons and other lower-energy secondary emissions) that is capable of facile tumor destruction. Since Rose Bengal will tend to concentrate in cellular membranes, the released energy for such targeted material will be ideally suited for stimulation of cell necrosis as a consequence of acute membrane damage resulting from interaction of the therapeutic energy with such structures. Further, radiosensitizer agent present in the cytoplasm will be suitably disposed so as to facilitate damage to cellular genetic material and other cellular structures as a consequence of similar secondary emission mechanisms. Further, because the radiosensitizer and radioisotope is rapidly cleared from the body, the complexity of any necessary follow-up procedure is minimal, since the body will tend to eliminate residual radiosensitizer, radioactive material, and destroyed tumor products via natural means.

This description has been offered for illustrative purposes only and is not intended to limit the invention of this application, which is defined in the claims below. For example, it will be clear to those of ordinary skill in the art that the targeting described herein for the specific example of the halogenated xanthenes can be adapted or otherwise applied to other radiodense materials, including conventional radiosensitizers.

Accordingly, the scope of the present invention is not as described in detail above, but as set forth in the appended claims.

We claim:

1. A method for treating a selected volume of tissue, said method comprising:  
distributing a radiosensitizer within said volume; and  
distributing a plurality of ionizing radiation sources within said volume, each  
radiation source producing a radiation treatment zone, and substantially all of the tissue  
in said tissue volume residing within at least one radiation treatment zone.

2. The method of Claim 1 in which said radiosensitizer is substantially  
uniformly distributed within said volume.

3. The method of Claim 1 in which said radiation sources are distributed  
within said volume of tissue such that said treatment zones are generally uniformly  
distributed throughout said volume of tissue.

4. The method of Claim 1 wherein each of said treatment zones overlaps at  
least one other treatment zone.

5. The method of Claim 1 wherein said radiosensitizer and said radiation  
sources are joined to define a conjugate agent.

6. The method of Claim 1 wherein said radiosensitizer is joined to a targeting  
moiety to define a conjugate agent and said radiation sources are joined to a targeting  
moiety to define another conjugate agent.

7. The method of Claim 1 wherein said radiosensitizer and said radiation  
sources are located in a delivery vehicle to define an encapsulated agent.

8. The method of Claim 5 wherein said distributing of a radiosensitizer and  
said distributing of ionizing radiation sources are carried out simultaneously by injecting  
said conjugate agent into or proximal to said volume of tissue.

9. The method of Claim 1 wherein said distributing of a radiosensitizer and  
said distributing of ionizing radiation sources are carried out by sequentially administering  
said radiosensitizer and said ionizing radiation sources.

10. The method of Claim 1 wherein said radiosensitizer, when distributed, conjugates with said tissue.

5 11. The method of Claim 1 wherein said radiation source, when distributed, conjugates with said tissue.

12. The method of Claim 1 wherein said treatment zones do not extend substantially beyond said selected volume of tissue.

10 13. The method of Claim 1 further comprising introducing said radiosensitizers systemically.

14. The method of Claim 1 further comprising introducing said radiosensitizers directly into said selected volume of tissue.

15 15. The method of Claim 14 wherein said distributing occurs after said introducing.

20 16. The method of Claim 14 wherein said distributing occurs during said introducing.

17. The method of Claim 1 further comprising injecting said radiosensitizers into proximity of said selected volume of tissue.

25 18. The method of Claim 17 wherein said distributing occurs after said introducing.

30 19. The method of Claim 17 wherein said distributing occurs during said introducing.

20. The method of Claim 1 further comprising introducing said ionizing radiation sources directly into said selected volume of tissue.



21. The method of Claim 20 wherein said distributing occurs after said introducing.

5 22. The method of Claim 20 wherein said distributing occurs during said introducing.

23. The method of Claim 1 wherein said selected volume of tissue comprises tissue experiencing undue cell proliferation.

10 24. The method of Claim 1 wherein said radiosensitizers are substantially uniformly activated throughout said selected volume of tissue.

25. The method of Claim 1 wherein said ionizing radiation sources emit minimal effective radiation outside said selected volume of tissue.

15 26. The method of Claim 1 wherein ionizing radiation source is a tumor-dispersible radioactive material.

20 27. The method of Claim 26 wherein said tumor-dispersible radioactive material is selected from the group consisting of radioisotopes attached to or encompassed in organic or inorganic microspheres, micelles, nanoparticles and colloids, or solubilized using chelates, organic agents and inorganic agents.

25 28. The method of Claim 1 wherein said radiosensitizer is selected from the group consisting of radiodense halogenated xanthenes and their derivatives; x-ray contrast agents; agents containing radiodense elements; iododeoxyuridine and bromodeoxyuridine and related agents; halogenated nucleotides and DNA ligands and intercalators; substituted acridine and imidazole based agents; nitroimidazoles and related bioreductive agents; misonidazole and related agents; etanidazole and related agents; pimondazole and  
30 related piperidine-derivatives; aziridines and related agents; cyclophosphamide and related agents; nitrosoureas and related agents; L-phenylalanine mustard and related agents; cis-platinum compounds and related agents; and doxorubicin and related agents.

5 29. The method of Claim 28 wherein said halogenated xanthenes are selected from the group consisting of Rose Bengal and its derivatives, Phloxine B and its derivatives, Erythrosin B and its derivatives, Eosin Y and its derivatives, highly brominated halogenated xanthenes, and highly iodinated halogenated xanthenes and their derivatives, including 4,5,6,7-Tetrabromoerythrosin.

10 30. The method of Claim 28 wherein said x-ray contrast agents are selected from the group consisting of Omnipaque™ (iohexol), Omniscan™ (gadodiamide), WIN 8883 (diatrizoic acid), iodamide and lipiodol.

31. The method of Claim 28 wherein said radiodense elements are selected from the group consisting of iodine, bromine, chlorine, barium, bismuth, boron, gold, silver, platinum, iron, gadolinium, dysprosium, and tantalum.

15 32. The method of Claim 1 wherein said radiosensitizer has a concentration of between about 1 ng to 10 g of radiosensitizer per kg of tissue.

33. The method of Claim 1 wherein said ionizing radiation source provides a dose within said treatment zone of between about 1 mGy and 1000 Gy.

20 34. The method of Claim 33 wherein said dose is between about 0.1 Gy and 100 Gy.

25 35. The method of Claim 5 wherein said radiation source is a radioactive moiety selected from the group consisting of aluminum, americium, cobalt, copper, gallium, gold, indium, iodine, iridium, manganese, phosphorus, radium, rhenium, rhodium, ruthenium, sulfur, technetium thallium, yttrium, and zinc.

30 36. The method of Claim 6 wherein said targeting moiety is selected from the group consisting of DNA, RNA, amino acids, proteins, antibodies, ligands, haptens, carbohydrate receptors, protein receptors, lipid receptors, chelators, encapsulating vehicles, nanoparticles short- and long-chain aliphatic and aromatic hydrocarbons, and other hydrophilic or hydrophobic moieties, and complexing agents.

37. The method of Claim 36 wherein said short- and long-chain aliphatic and aromatic hydrocarbon include one or more compounds selected from the group consisting of aldehydes, ketones, alcohols, esters, amides, amines, nitriles, and azides.

38. The method of Claim 7 wherein said delivery vehicle is selected from the group consisting of a micelle, liposome and nanoparticle.

39. An agent for treatment of tissue, said agent comprising:  
a radiosensitizer component and  
an ionizing radiation source component,  
said radiosensitizer component and radiation source component being combined to define an injectable treatment agent.

40. The agent of Claim 39 wherein said agent is in liquid form.

41. The agent of Claim 39 wherein said agent is in gel form.

42. The agent of Claim 39 wherein said radiosensitizer component and radiation source component are conjugated.

43. The agent of Claim 39 wherein said radiosensitizer component and said radiation source component are located in a delivery vehicle to define an encapsulated agent.

44. The agent of claim 39 wherein ionizing radiation source component is a tumor-dispersible radioactive material.

45. The agent of Claim 44 wherein said tumor-dispersible radioactive material is selected from the group consisting of radioisotopes attached to or encompassed in organic or inorganic microspheres, micelles, nanoparticles and colloids, or solubilized using chelates, organic agents and inorganic agents.

46. The agent of Claim 39 wherein said radiosensitizer is selected from the group consisting of radiodense halogenated xanthenes and their derivatives; x-ray contrast agents; agents containing radiodense elements; iododeoxyuridine and bromodeoxyuridine and related agents; halogenated nucleotides and DNA ligands and intercalators; substituted acridine and imidazole based agents; nitroimidazoles and related bioreductive agents; misonidazole and related agents; etanidazole and related agents; pimondazole and related piperidine-derivatives; aziridines and related agents; cyclophosphamide and related agents; nitrosoureas and related agents; L-phenylalanine mustard and related agents; cis-platinum compounds and related agents; and doxorubicin and related agents.

47. The agent of Claim 46 wherein said halogenated xanthenes are selected from the group consisting of Rose Bengal and its derivatives, Phloxine B and its derivatives, Erythrosin B and its derivatives, Eosin Y and its derivatives, highly brominated halogenated xanthenes, and highly iodinated halogenated xanthenes and their derivatives, including 4,5,6,7-Tetrabromoerythrosin.

48. The agent of Claim 46 wherein said x-ray contrast agents are selected from the group consisting of Omnipaque™ (iohexol), Omniscan™ (gadodiamide), WIN 8883 (diatrizoic acid), iodamide and lipiodol.

49. The agent of Claim 46 wherein said radiodense elements are selected from the group consisting of iodine, bromine, chlorine, barium, bismuth, boron, gold, silver, platinum, iron, gadolinium, dysprosium, and tantalum.

50. The agent of Claim 39 wherein said radiosensitizer component has a concentration of between about 1 ng to 10 g of radiosensitizer per kg of tissue.

51. The agent of Claim 39 wherein said ionizing radiation source component provides a dose within said treatment zone of between about 1 mGy and 1000 Gy.

52. The agent of Claim 51 wherein said dose is between about 0.1 Gy and 100 Gy.

53. The agent of Claim 39 wherein radiation source component is a radioactive moiety selected from the group consisting of aluminum, americium, cobalt, copper, gallium, gold, indium, iodine, iridium, manganese, phosphorus, radium, rhenium, rhodium, ruthenium, sulfur, technetium, thallium, yttrium, and zinc.

54. The agent of Claim 39 wherein said radiosensitizer component and said radiation source component are located in a delivery vehicle to define an encapsulated agent.

55. The agent of Claim 54 wherein said delivery vehicle is selected from the group consisting of a micelle, liposome, and nanoparticle.

56. An agent For treatment of tissue, said agent comprising:  
a conjugate agent comprised of a radiosensitizer jointed to a targeting moiety; and  
a second conjugate agent comprised of a radiation source jointed to a targeting moiety.

57. The agent of Claim 56 wherein said targeting moiety is selected from the group consisting of DNA, RNA, amino acids, proteins, antibodies, ligands, haptens, carbohydrate receptors, protein receptors, lipid receptors chelators, encapsulating vehicles, nanoparticles short- and long-chain aliphatic and aromatic hydrocarbons, and other hydrophilic or hydrophobic moieties, and complexing agents.

58. The agent of Claim 57 wherein said short- and long-chain aliphatic and aromatic hydrocarbon include one or more compounds selected from the group consisting of aldehydes, ketones, alcohols, esters, amides, amines, nitriles, and azides.

59. The agent of Claim 56 wherein ionizing radiation source component is a tumor-dispersible radioactive material.

60. The agent of Claim 59 wherein said tumor-dispersible radioactive material is selected from the group consisting of radioisotopes attached to or encompassed in

organic or inorganic microspheres, micelles, nanoparticles and colloids, or solubilized using chelates, organic agents and inorganic agents.

5           61.    The agent of Claim 56 wherein said radiosensitizer is selected from the group consisting of radiodense halogenated xanthenes and their derivatives; x-ray contrast agents; agents containing radiodense elements; iododeoxyuridine and bromodeoxyuridine and related agents; halogenated nucleotides and DNA ligands and intercalators; substituted acridine and imidazole based agents; nitroimidazoles and related bioreductive agents; misonidazole and related agents; etanidazole and related agents; pimondazole and  
10   related piperidine-derivatives; aziridines and related agents; cyclophosphamide and related agents; nitrosoureas and related agents; L-phenylalanine mustard and related agents; cis-platinum compounds and related agents; and doxorubicin and related agents.

15           62.    The agent of Claim 61 wherein said halogenated xanthenes are selected from the group consisting of Rose Bengal and its derivatives, Phloxine Ba nd its derivatives, Erythrosin B and its derivatives, Eosin Y and its derivatives, highly brominated halogenated xanthenes, and highly iodinated halogenated xanthenes and their derivatives, including 4,5,6,7-Tetrabromoerythrosin.

20           63.    The agent of Claim 61 wherein said x-ray contrast agents are selected from the group consisting of Omnipaque™ (iohexol), Omniscan™ (gadodiamide), WIN 8883 (diatrizoic acid), iodamide and lipiodol.

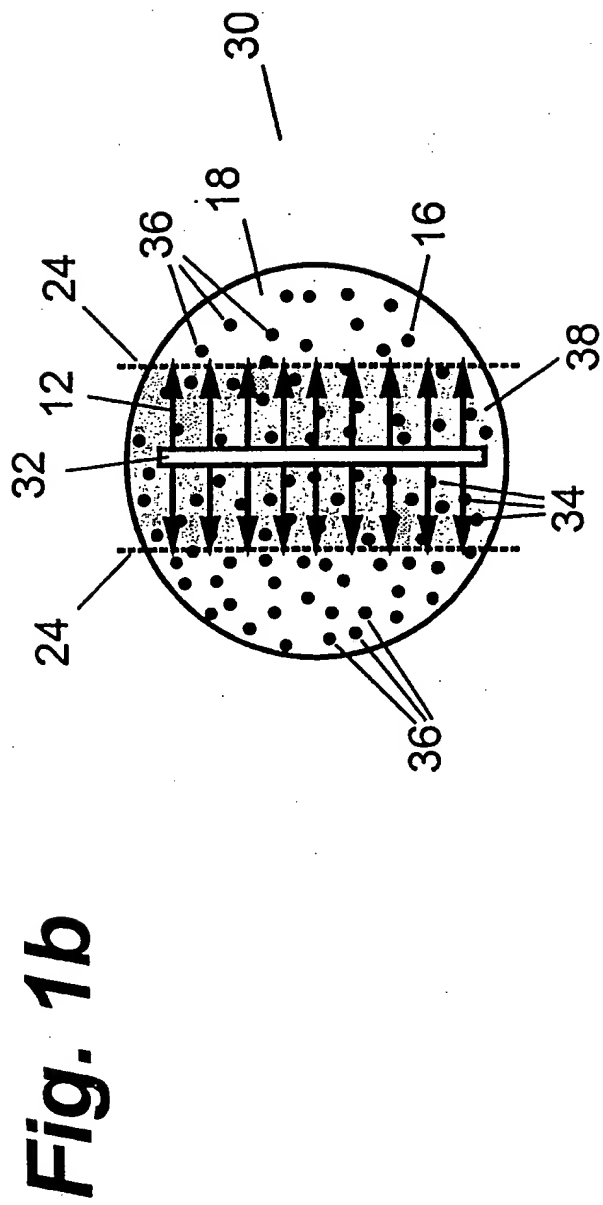
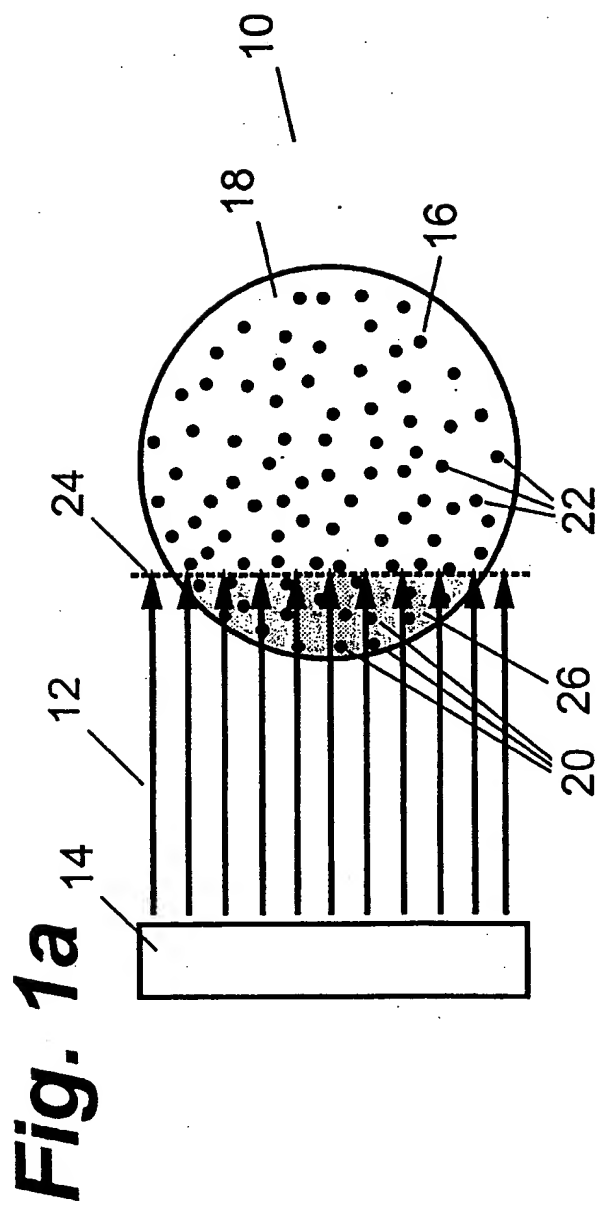
25           64.    The agent of Claim 61 wherein said radiodense elements are selected from the group consisting of iodine, bromine, chlorine, barium, bismuth, boron, gold, silver, platinum, iron, gadolinium, dysprosium, and tantalum.

30           65.    The agent of Claim 56 wherein said radiation source component is a radioactive moiety selected from the group consisting of aluminum, americium, cobalt, copper, gallium, gold, indium, iodine, iridium, manganese, phosphorus, radium, rhenium, rhodium, ruthenium, sulfur, technetium, thallium, yttrium, and zinc.

66. The method of Claim 1 wherein said distributing of a radiosensitizer and said distributing of ionizing radiation sources are carried out by simultaneously administering said radiosensitizer and said ionizing radiation sources.

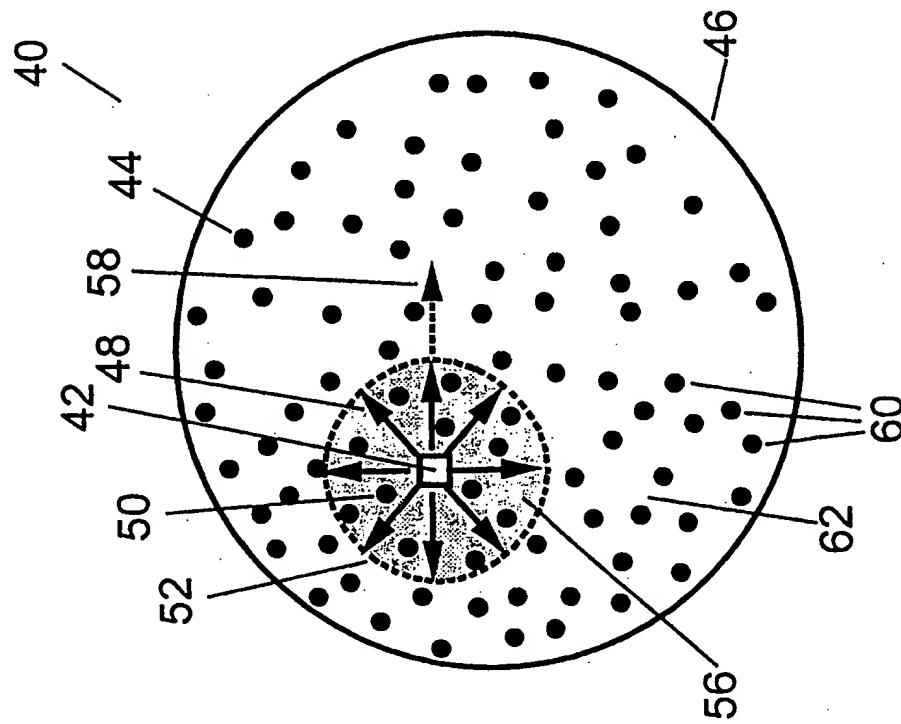
5 67. A method for treating a selected volume of tissue comprising the steps of:  
administering a radiosensitizer to a patient, a portion of said radiosensitizer being  
retained substantially throughout said volume of tissue; and

10 locally administering a plurality of radiation sources in proximity to or into said  
volume of tissue, said radiation sources distributing through said volume of tissue to  
produce a substantially uniform radiation throughout said volume of tissue.

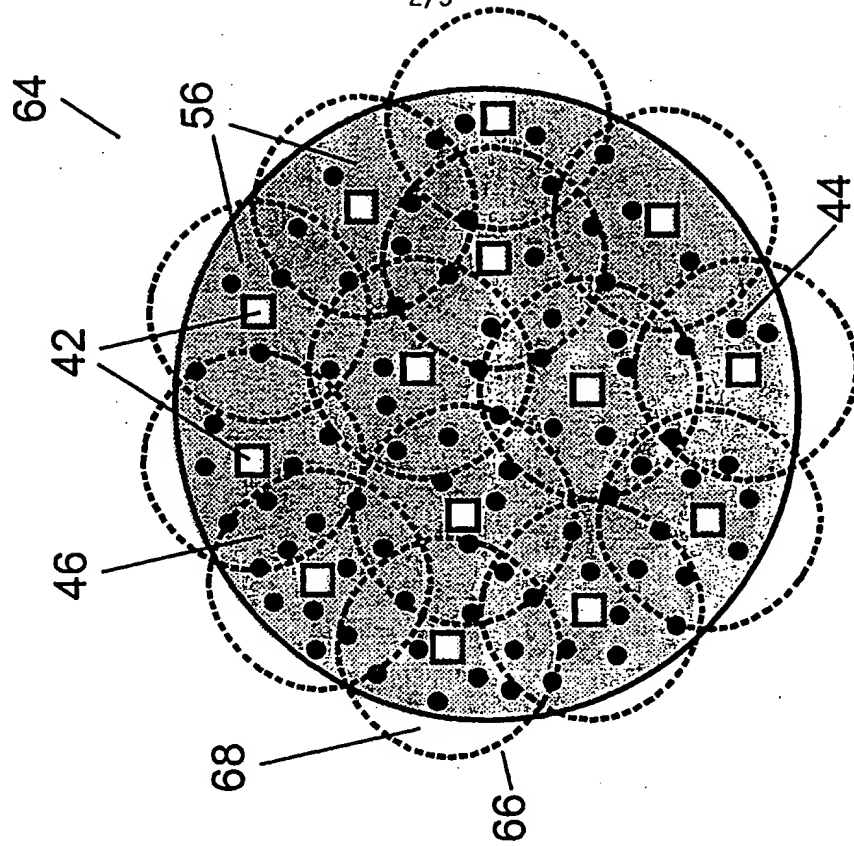




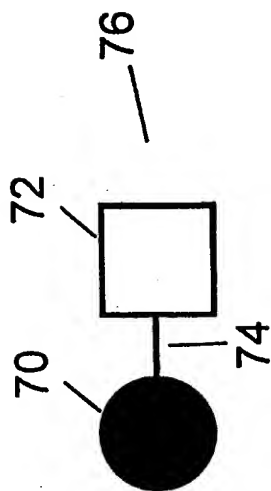
**Fig. 2a**



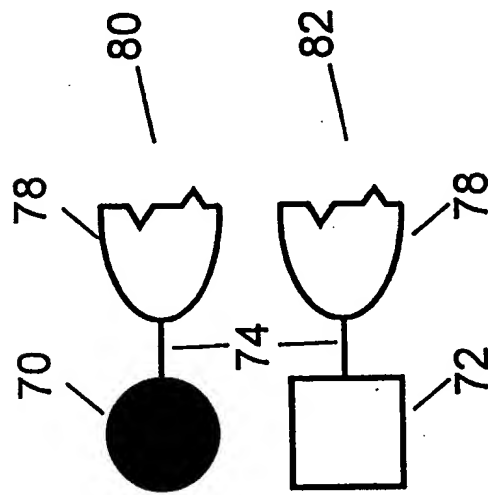
**Fig. 2b**



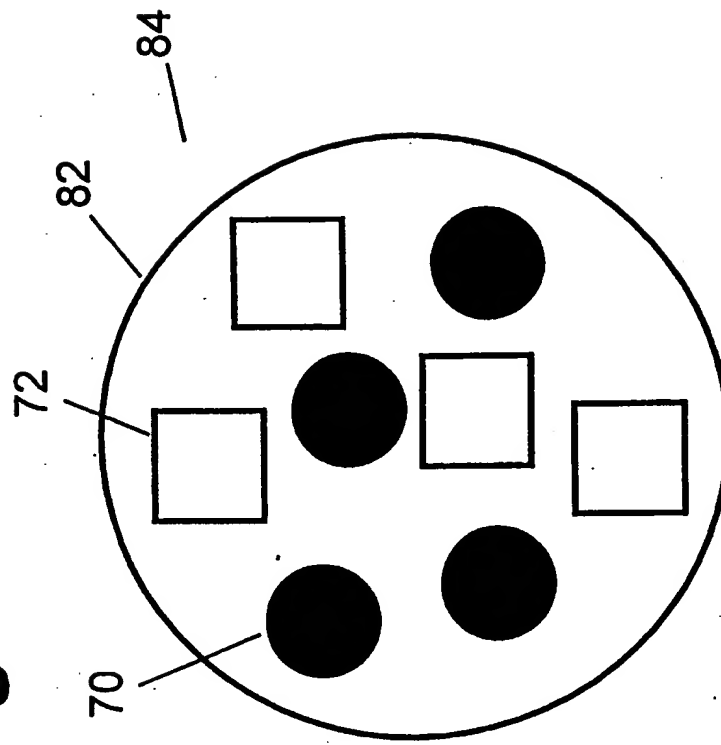
**Fig. 3a**



**Fig. 3b**



**Fig. 3c**



# INTERNATIONAL SEARCH REPORT

International application No.

PCT/US00/01815

## A. CLASSIFICATION OF SUBJECT MATTER

IPC : A61K 51/00

US CL : 424/1.33

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 424/1.33

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
WEST, STN

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category * | Citation of document, with indication, where appropriate, of the relevant passages  | Relevant to claim No.   |
|------------|---|---|
| X, P       | US 6,015,541 A (GREFF et al) 18 January 2000 (18-01-00) columns 3-5, 7, 11.   | 1-28, 30-31, 33-37, 39-46, 48-49, 51-54, 56-61, 63-67           |
| X          | US 5,342,283 A (GOOD) 30 August 1994 (30-08-94) columns 12, 15, 45-47, 49-50.   | 1-28, 31, 33-37, 39, 42-46, 49, 51-54, 56-61, 64-67             |
| X          | MORSTYN et al. 131-Iodine Conjugated Antibody Cell Kill Enhanced by Bromodeoxyuridine. Int. J. Radiation Oncology Biol. Phys. August 1984, Vol. 10, No. 8, pages 1437-1440, see discussion section. | 1-6, 8-12, 14-28, 31, 34-36, 39, 42, 44-46, 52-53, 56-61, 64-67 |

☐ Further documents are listed in the continuation of Box C.

☐ See patent family annex.

\* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T"

later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X"

document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y"

document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&"

document member of the same patent family

Date of the actual completion of the international search

05 April 2000 (05.04.2000)

Date of mailing of the international search report

24 MAY 2000

Name and mailing address of the ISA/US

Commissioner of Patents and Trademarks  
Box PCT  
Washington, D.C. 20231

Facsimile No. (703) 305-3230

Authorized officer

Gary Nickol

Telephone No. (703) 308-0196